The Role of M1BP in Eye Development of *Drosophila melanogaster*

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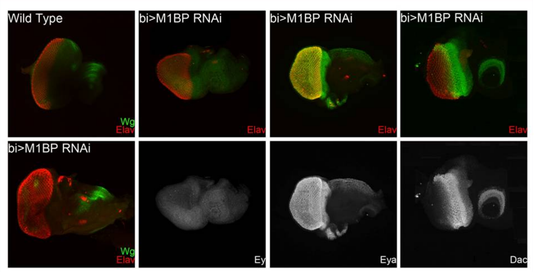
Abstract

Many genes in the *Drosophila melanogaster* have Pol II paused at the promoter proximal region, because the binding of either the GAGA factor or the Motif 1 binding protein (M1BP). M1BP resides on chromosome 2 of *Drosophila melanogaster* and directs a distinct transcriptional mechanism evolved from the TATA box. M1BP is highly conserved across the species and encodes a 55kDa protein containing five C2H2 zinc-fingers domains. A battery of highly conserved genes regulates drosophila eye development. Based on high throughput studies, it has been suggested that M1BP may regulate gene expression during Drosophila eye development, but its exact role is unknown. Our aim is to study the role of M1BP during eye development. We have used Green Fluorescent Protein (GFP) marker to identify intended regions to be expressed. This GFP marker has expressed the dorsal, ventral, morphogenetic furrow and the entire eye. This aim is further focused with absence of M1BP being produced in the stock fly and then focusing on the phenotype and genotype when crossed with another set of flies that have a suppression in development of some aspect of the eye. We found that absence of M1BP function in dorsal and ventral eye margins results in the suppression of eye fate. This suppression of eye fate was found when both the dorsal and ventral margins were expressed, along with a suppression of eye fate when the dorsal and ventral regions were expressed separately. The absence of M1BP also led to the suppression of the gene from the complete eye, giving us a head loss phenotype. This head loss phenotype shows the destruction and absence of photoreceptors in the developmental stages of the eye.

Motif 1 Binding Protein (M1BP)

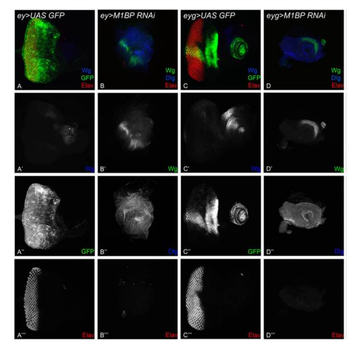
- Novel Transcription Factor, which has one annotated transcript and one polypeptide
- Encodes a 55KDa Protein containing five C2H2 zinc-fingers domains
- Molecular function:
  - Zinc ion binding
  - RNA polymerase II core promoter sequence specific DNA binding
- Binds to the promoters of paused genes and controls their expressions

Retinal determination genes expression extends into the anterior head cuticular region



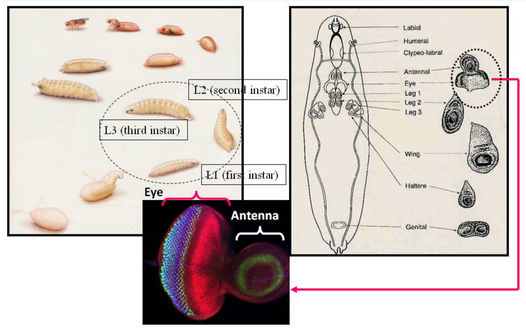
Ey is downregulated, Eya and Dac moves anteriorly into the region that marks the retinal precursor cells.

Role of M1BP RNAi in Eye Development

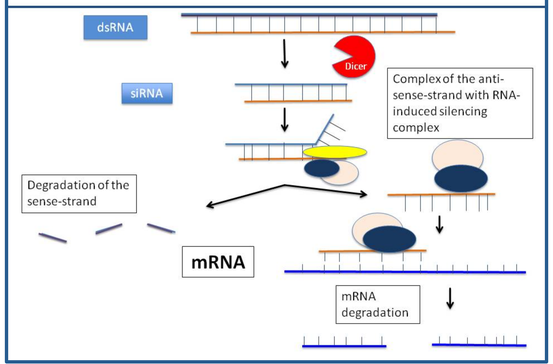


Drosophila has a short life cycle of 12 days.

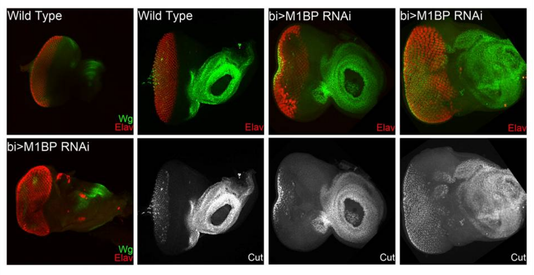
Imaginal disc housed in the larva serves as blue print for adult organs.



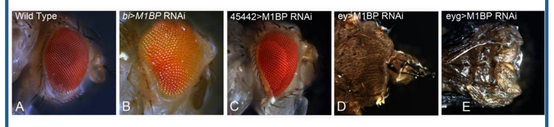
RNA Interference (RNAi)



Antenna specific marker Cut expression is induced in region anterior to Morphogenetic Furrow

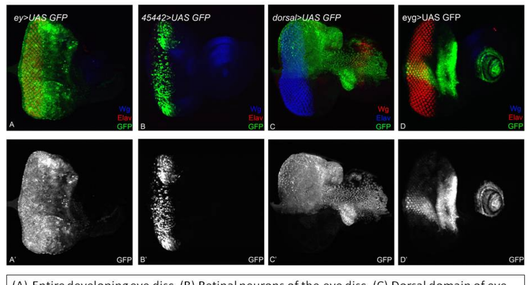


Loss of M1BP on DV margin, Ventral, entire eye and equatorial region of eye results in reduced eye and "no-eye" phenotype



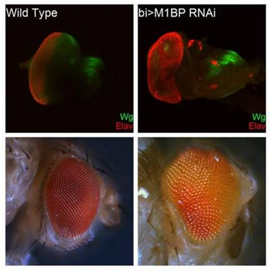
Loss of M1BP function in the eye. (A) Wild type eye. (B) Suppression of eye fate in the absence of M1BP function from the dorsal and ventral margins of the eye. (C) Suppression of eye fate in the absence of M1BP function from ventral margins of the eye. (D,E) Headless adult phenotype in the absence of M1BP function from the entire eye.

GFP reporter marks the domains of developing eye disc utilized by the Gal4-UAS target system



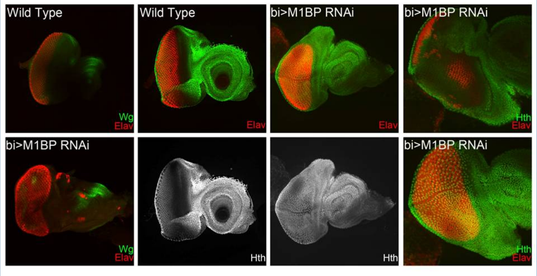
(A) Entire developing eye disc. (B) Retinal neurons of the eye disc. (C) Dorsal domain of eye disc. (D) Equatorial extending to the optic stalk.

Loss of MBP1 results in change of peripodial epithelium to disc proper retinal fate



Ectopic Wg expression is seen on new disc margin that folds into PE.

Hth expression, a negative regulator of eye development, is modified



A. Hth expression in PE is downregulated near PE and DP border
B. Hth anterior to MF also moves more towards anterior margin.

Conclusion

- M1BP is required to define eye versus head boundary
- Loss of M1BP results in change of peripodial epithelium fate to the disc proper fate.
- Loss of M1BP function in the eye gives a headless adult phenotype
- Loss of M1BP function in eye shows photoreceptors in disarray during larva stage
- The mechanism for M1BP mediated regulation of eye specific fate is yet to be determined

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